



UM01531.ST25.txt
SEQUENCE LISTING

<110> Hale, Calvin C
Price, Elmer M

<120> LARGE SCALE EXPRESSION AND PURIFICATION OF RECOMBINANT PROTEINS

<130> UMO 1531.1

<140> US 09/901,419

<141> 2001-07-09

<160> 5

<170> PatentIn version 3.1

C3

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<211> 4087

<212> DNA

<213> Bos taurus

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<222> (268)..(3180)

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<221> sig_peptide

<222> (268)..(363)

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<221> misc_feature

<222> (3178)..()

<223> A Poly (H) affinity tag comprising 6 His residues have been inserted at the C-Terminus end of the coding region of the protein

<400> 1

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cagagtctcg agatgcgacg ctacaatctg cagttttcac tagcttccca gtaggttggg      240
acagttggaa ctctgccatt gcccagc  atg ctg cag ttc agt ctg tca ccc acc      294
                               Met Leu Gln Phe Ser Leu Ser Pro Thr
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Leu Ser Met Gly Phe His Val Ile Ala Met Val Ala Leu Leu Phe Ser
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Glu Thr Gly Glu Cys Thr Gly Ser Tyr Tyr Cys Lys Lys Gly Val Ile
                               45                               50                               55

tta ccc att tgg gag ccc cag gac cct tcc ttt gga gac aaa att gct      486
Leu Pro Ile Trp Glu Pro Gln Asp Pro Ser Phe Gly Asp Lys Ile Ala
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Arg Ala Thr Val Tyr Phe Val Ala Met Val Tyr Met Phe Leu Gly Val
75                               80                               85

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Ser Ile Ile Ala Asp Arg Phe Met Ser Ser Ile Glu Val Ile Thr Ser
90                               95                               100                               105

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Gln Glu Lys Glu Ile Thr Ile Lys Lys Pro Asn Gly Glu Thr Thr Lys
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125                               130                               135

gcc ctg ggg tct tca gct cca gag att ctc ctt tca gta atc gag gtg      726
Ala Leu Gly Ser Ser Ala Pro Glu Ile Leu Leu Ser Val Ile Glu Val
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ctt act ttc ttc ttc ttc ccc atc tgc gtt gtg ttt gct tgg gtg gca Leu Thr Phe Phe Phe Phe Pro Ile Cys Val Val Phe Ala Trp Val Ala 235 240 245	1014
gac agg agg ctt ctg ttt tac aag tat gtc tac aag agg tat cgg gct Asp Arg Arg Leu Leu Phe Tyr Lys Tyr Val Tyr Lys Arg Tyr Arg Ala 250 255 260 265	1062
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aac acg gaa gtg gct gaa aat gac cct gtc agt aag atc ttc ttt gaa Asn Thr Glu Val Ala Glu Asn Asp Pro Val Ser Lys Ile Phe Phe Glu 395 400 405	1494
caa ggg aca tat cag tgt ctg gag aac tgt ggc aca gta gcc ctg acc Gln Gly Thr Tyr Gln Cys Leu Glu Asn Cys Gly Thr Val Ala Leu Thr 410 415 420 425	1542

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gaa gga act gtg gtc ttt aag cct ggt gag acc cag aag gaa atc aga Glu Gly Thr Val Val Phe Lys Pro Gly Glu Thr Gln Lys Glu Ile Arg 460 465 470	1686
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Phe																	
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<213> Bos taurus

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<222> (3178)..()

<223> A Poly (H) affinity tag comprising 6 His residues have been inser

ted at the C-Terminus end of the coding region of the protein

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Ser Tyr Tyr Cys Lys Lys Gly Val Ile Leu Pro Ile Trp Glu Pro Gln
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Asp Pro Ser Phe Gly Asp Lys Ile Ala Arg Ala Thr Val Tyr Phe Val
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Ala Met Val Tyr Met Phe Leu Gly Val Ser Ile Ile Ala Asp Arg Phe
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Met Ser Ser Ile Glu Val Ile Thr Ser Gln Glu Lys Glu Ile Thr Ile
100 105 110

Lys Lys Pro Asn Gly Glu Thr Thr Lys Thr Thr Val Arg Ile Trp Asn
115 120 125

Glu Thr Val Ser Asn Leu Thr Leu Met Ala Leu Gly Ser Ser Ala Pro
130 135 140

Glu Ile Leu Leu Ser Val Ile Glu Val Cys Gly His Asn Phe Thr Ala
145 150 155 160

Gly Asp Leu Gly Pro Ser Thr Ile Val Gly Ser Ala Ala Phe Asn Met
165 170 175

Phe Ile Ile Ile Ala Leu Cys Val Tyr Val Val Pro Asp Gly Glu Thr
180 185 190

Arg Lys Ile Lys His Leu Arg Val Phe Phe Val Thr Ala Ala Trp Ser
195 200 205

Ile Phe Ala Tyr Thr Trp Leu Tyr Ile Ile Leu Ser Val Ser Ser Pro
210 215 220

Gly Val Val Glu Val Trp Glu Gly Leu Leu Thr Phe Phe Phe Phe Pro
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260 265 270

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275 280 285

Asp Gly Lys Val Val Asn Ser His Val Asp Ser Phe Leu Asp Gly Ala
290 295 300

Leu Val Leu Glu Val Asp Glu Arg Asp Gln Asp Asp Glu Glu Ala Arg
305 310 315 320

Arg Glu Met Ala Arg Ile Leu Lys Glu Leu Lys Gln Lys His Pro Glu
325 330 335

Lys Glu Ile Glu Gln Leu Ile Glu Leu Ala Asn Tyr Gln Val Leu Ser
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Gln Gln Gln Lys Ser Arg Ala Phe Tyr Arg Ile Gln Ala Thr Arg Leu
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Met Thr Gly Ala Gly Asn Ile Leu Lys Arg His Ala Ala Asp Gln Ala
370 375 380

Arg Lys Ala Val Ser Met His Glu Val Asn Thr Glu Val Ala Glu Asn
385 390 395 400

Asp Pro Val Ser Lys Ile Phe Phe Glu Gln Gly Thr Tyr Gln Cys Leu
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Glu Asn Cys Gly Thr Val Ala Leu Thr Ile Ile Arg Arg Gly Gly Asp
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Leu Thr Asn Thr Val Phe Val Asp Phe Arg Thr Glu Asp Gly Thr Ala
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Asn Ala Gly Ser Asp Tyr Glu Phe Thr Glu Gly Thr Val Val Phe Lys
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Pro Gly Glu Thr Gln Lys Glu Ile Arg Val Gly Ile Ile Asp Asp Asp
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Ile Phe Glu Glu Asp Glu Asn Phe Leu Val His Leu Ser Asn Val Lys
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Val Ser Leu Glu Ala Ser Glu Asp Gly Ile Leu Glu Ala Ser His Val
500 505 510

Ser Thr Leu Ala Cys Leu Gly Ser Pro Ser Thr Ala Thr Val Thr Ile
515 520 525

Phe Asp Asp Asp His Ala Gly Ile Phe Thr Phe Glu Glu Pro Val Thr
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His Val Ser Glu Ser Ile Gly Ile Met Glu Val Lys Val Leu Arg Thr
545 550 555 560

Ser Gly Ala Arg Gly Asn Val Ile Val Pro Tyr Lys Thr Ile Glu Gly
565 570 575

Thr Ala Arg Gly Gly Gly Glu Asp Phe Glu Asp Thr Cys Gly Glu Leu
580 585 590

Glu Phe Gln Asn Asp Glu Ile Val Lys Thr Ile Ser Val Lys Val Ile
595 600 605

Asp Asp Glu Glu Tyr Glu Lys Asn Lys Thr Phe Phe Leu Glu Ile Gly
610 615 620

Glu Pro Arg Leu Val Glu Met Ser Glu Lys Lys Ala Leu Leu Leu Asn
625 630 635 640

Glu Leu Gly Gly Phe Thr Ile Thr Gly Lys Tyr Leu Tyr Gly Gln Pro
645 650 655

Val Phe Arg Lys Val His Ala Arg Glu His Pro Leu Pro Ser Thr Ile
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Ile Thr Ile Ala Asp Glu Tyr Asp Asp Lys Gln Pro Leu Thr Ser Lys
675 680 685

Glu Glu Glu Glu Arg Arg Ile Ala Glu Met Gly Arg Pro Ile Leu Gly
690 695 700

Glu His Thr Arg Leu Glu Val Ile Ile Glu Glu Ser Tyr Glu Phe Lys
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Ser Thr Val Asp Lys Leu Ile Lys Lys Thr Asn Leu Ala Leu Val Val
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Gly Thr Asn Ser Trp Arg Glu Gln Phe Ile Glu Ala Ile Thr Val Ser

C3
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740

745

750

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 770 775 780

Leu Phe Ala Phe Val Pro Pro Thr Glu Tyr Trp Asn Gly Trp Ala Cys
 785 790 795 800

Phe Ile Val Ser Ile Leu Met Ile Gly Leu Leu Thr Ala Phe Ile Gly
 805 810 815

Asp Leu Ala Ser His Phe Ala Cys Thr Ile Ala Leu Lys Asp Ser Val
 820 825 830

Thr Ala Val Val Phe Val Ala Leu Gly Thr Ser Val Pro Asp Thr Phe
 835 840 845

Ala Ser Lys Val Ala Ala Thr Gln Asp Gln Tyr Ala Asp Ala Ser Ile
 850 855 860

Gly Asn Val Thr Gly Ser Asn Ala Val Asn Val Phe Leu Gly Ile Gly
 865 870 875 880

Val Ala Trp Ser Ile Ala Ala Ile Tyr His Ala Ala Asn Gly Glu Gln
 885 890 895

Phe Lys Val Ser Pro Gly Thr Leu Ala Phe Ser Val Thr Leu Phe Thr
 900 905 910

Ile Phe Ala Phe Ile Asn Val Gly Val Leu Leu Tyr Arg Arg Arg Pro
 915 920 925

Glu Ile Gly Gly Glu Leu Gly Gly Pro Arg Thr Ala Lys Leu Leu Thr
 930 935 940

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<213> Bos taurus

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Glu Thr Glu Met Glu Gly Glu Gly Asn Glu Thr Gly Glu Cys Thr Gly
 35          40          45

Ser Tyr Tyr Cys Lys Lys Gly Val Ile Leu Pro Ile Trp Glu Pro Gln
 50          55          60

Asp Pro Ser Phe Gly Asp Lys Ile Ala Arg Ala Thr Val Tyr Phe Val
 65          70          75          80

Ala Met Val Tyr Met Phe Leu Gly Val Ser Ile Ile Ala Asp Arg Phe
 85          90          95

Met Ser Ser Ile Glu Val Ile Thr Ser Gln Glu Lys Glu Ile Thr Ile
100          105          110

Lys Lys Pro Asn Gly Glu Thr Thr Lys Thr Thr Val Arg Ile Trp Asn
115          120          125

Glu Thr Val Ser Asn Leu Thr Leu Met Ala Leu Gly Ser Ser Ala Pro
130          135          140

Glu Ile Leu Leu Ser Val Ile Glu Val Cys Gly His Asn Phe Thr Ala
145          150          155          160

Gly Asp Leu Gly Pro Ser Thr Ile Val Gly Ser Ala Ala Phe Asn Met
165          170          175

Phe Ile Ile Ile Ala Leu Cys Val Tyr Val Val Pro Asp Gly Glu Thr
180          185          190

Arg Lys Ile Lys His Leu Arg Val Phe Phe Val Thr Ala Ala Trp Ser
195          200          205

Ile Phe Ala Tyr Thr Trp Leu Tyr Ile Ile Leu Ser Val Ser Ser Pro
210          215          220

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Gly Val Val Glu Val Trp Glu Gly Leu Leu Thr Phe Phe Phe Phe Pro
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Ile Cys Val Val Phe Ala Trp Val Ala Asp Arg Arg Leu Leu Phe Tyr
245 250 255

Lys Tyr Val Tyr Lys Arg Tyr Arg Ala Gly Lys Gln Arg Gly Met Ile
260 265 270

Ile Glu His Glu Gly Asp Arg Pro Ser Ser Lys Thr Glu Ile Glu Met
275 280 285

Asp Gly Lys Val Val Asn Ser His Val Asp Ser Phe Leu Asp Gly Ala
290 295 300

Leu Val Leu Glu Val Asp Glu Arg Asp Gln Asp Asp Glu Glu Ala Arg
305 310 315 320

Arg Glu Met Ala Arg Ile Leu Lys Glu Leu Lys Gln Lys His Pro Glu
325 330 335

Lys Glu Ile Glu Gln Leu Ile Glu Leu Ala Asn Tyr Gln Val Leu Ser
340 345 350

Gln Gln Gln Lys Ser Arg Ala Phe Tyr Arg Ile Gln Ala Thr Arg Leu
355 360 365

Met Thr Gly Ala Gly Asn Ile Leu Lys Arg His Ala Ala Asp Gln Ala
370 375 380

Arg Lys Ala Val Ser Met His Glu Val Asn Thr Glu Val Ala Glu Asn
385 390 395 400

Asp Pro Val Ser Lys Ile Phe Phe Glu Gln Gly Thr Tyr Gln Cys Leu
405 410 415

Glu Asn Cys Gly Thr Val Ala Leu Thr Ile Ile Arg Arg Gly Gly Asp
420 425 430

Leu Thr Asn Thr Val Phe Val Asp Phe Arg Thr Glu Asp Gly Thr Ala
435 440 445

Asn Ala Gly Ser Asp Tyr Glu Phe Thr Glu Gly Thr Val Val Phe Lys
450 455 460

Pro Gly Glu Thr Gln Lys Glu Ile Arg Val Gly Ile Ile Asp Asp Asp
465 470 475 480

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Ile Phe Glu Glu Asp Glu Asn Phe Leu Val His Leu Ser Asn Val Lys
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Val Ser Leu Glu Ala Ser Glu Asp Gly Ile Leu Glu Ala Ser His Val
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Ser Thr Leu Ala Cys Leu Gly Ser Pro Ser Thr Ala Thr Val Thr Ile
515 520 525

Phe Asp Asp Asp His Ala Gly Ile Phe Thr Phe Glu Glu Pro Val Thr
530 535 540

His Val Ser Glu Ser Ile Gly Ile Met Glu Val Lys Val Leu Arg Thr
545 550 555 560

Ser Gly Ala Arg Gly Asn Val Ile Val Pro Tyr Lys Thr Ile Glu Gly
565 570 575

Thr Ala Arg Gly Gly Gly Glu Asp Phe Glu Asp Thr Cys Gly Glu Leu
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Glu Phe Gln Asn Asp Glu Ile Val Lys Thr Ile Ser Val Lys Val Ile
595 600 605

Asp Asp Glu Glu Tyr Glu Lys Asn Lys Thr Phe Phe Leu Glu Ile Gly
610 615 620

Glu Pro Arg Leu Val Glu Met Ser Glu Lys Lys Ala Leu Leu Leu Asn
625 630 635 640

Glu Leu Gly Gly Phe Thr Ile Thr Gly Lys Tyr Leu Tyr Gly Gln Pro
645 650 655

Val Phe Arg Lys Val His Ala Arg Glu His Pro Leu Pro Ser Thr Ile
660 665 670

Ile Thr Ile Ala Asp Glu Tyr Asp Asp Lys Gln Pro Leu Thr Ser Lys
675 680 685

Glu Glu Glu Glu Arg Arg Ile Ala Glu Met Gly Arg Pro Ile Leu Gly
690 695 700

Glu His Thr Arg Leu Glu Val Ile Ile Glu Glu Ser Tyr Glu Phe Lys
705 710 715 720

Ser Thr Val Asp Lys Leu Ile Lys Lys Thr Asn Leu Ala Leu Val Val

725

730

735

Gly Thr Asn Ser Trp Arg Glu Gln Phe Ile Glu Ala Ile Thr Val Ser
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Ala Gly Glu Asp Asp Asp Asp Asp Glu Cys Gly Glu Glu Lys Leu Pro
755 760 765

Ser Cys Phe Asp Tyr Val Met His Phe Leu Thr Val Phe Trp Lys Val
770 775 780

Leu Phe Ala Phe Val Pro Pro Thr Glu Tyr Trp Asn Gly Trp Ala Cys
785 790 795 800

Phe Ile Val Ser Ile Leu Met Ile Gly Leu Leu Thr Ala Phe Ile Gly
805 810 815

Asp Leu Ala Ser His Phe Ala Cys Thr Ile Ala Leu Lys Asp Ser Val
820 825 830

Thr Ala Val Val Phe Val Ala Leu Gly Thr Ser Val Pro Asp Thr Phe
835 840 845

Ala Ser Lys Val Ala Ala Thr Gln Asp Gln Tyr Ala Asp Ala Ser Ile
850 855 860

Gly Asn Val Thr Gly Ser Asn Ala Val Asn Val Phe Leu Gly Ile Gly
865 870 875 880

Val Ala Trp Ser Ile Ala Ala Ile Tyr His Ala Ala Asn Gly Glu Gln
885 890 895

Phe Lys Val Ser Pro Gly Thr Leu Ala Phe Ser Val Thr Leu Phe Thr
900 905 910

Ile Phe Ala Phe Ile Asn Val Gly Val Leu Leu Tyr Arg Arg Arg Pro
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Glu Ile Gly Gly Glu Leu Gly Gly Pro Arg Thr Ala Lys Leu Leu Thr
930 935 940

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